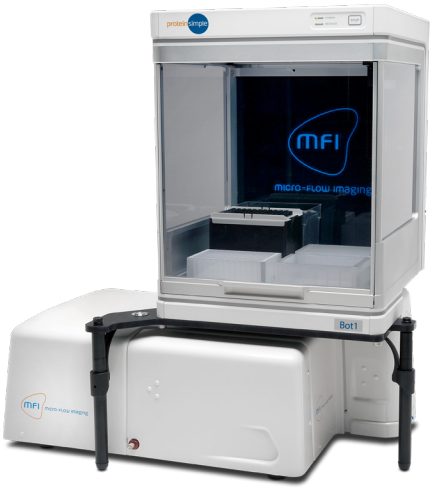
Image Analysis Software: New Features to Streamline Your Particle Characterization Workflow

Introduction

Micro-Flow Imaging™ (MFI) lets you capture images of sub-visible particles in your biopharmaceutical formulations by combining the direct imaging capabilities of digital microscopy with the precise control of a microfluidic approach. This insight is an important step in the regulatory approval process because sub-visible particles such as protein aggregates and/or process-related contaminants can cause safety and efficacy concerns, risking approval. With MFI, you'll get high-resolution images with 85% sampling efficiency. Your counts will be more precise, and you can size sub-particles with full morphological detail, giving you complete confidence that you've correctly identified every type possibly present.

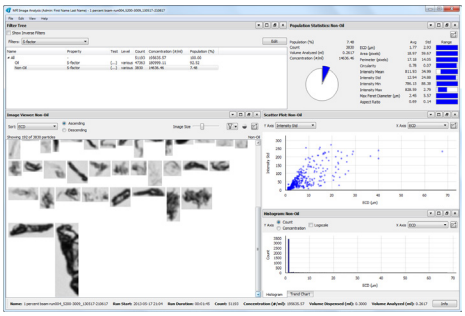


As an image-based technique, having the software in place to perform suitable data analysis is paramount. ProteinSimple's new MFI Image Analysis Software features improved analysis workflows and a new set of user interface options to give you more tools and increased flexibility when working with your particle data. In this technical note, we'll cover some of these new features and show you where you can find them within Image Analysis Software.

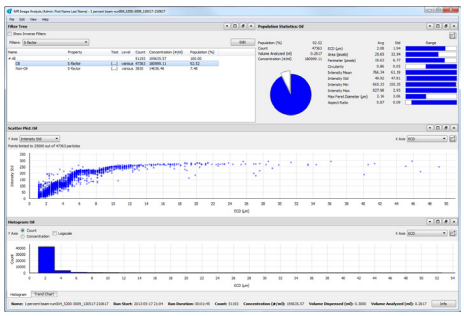
What's in the New User Interface?

MFI Image Analysis Software features a fully resizable and customizable interface (**Figure 1**). You can drag out and resize windows to take advantage of multiple monitors or dive deeper into the details of charts and images. Everything is at your fingertips, streamlining the data analysis to conclusion-making process.

A.



B.



C.

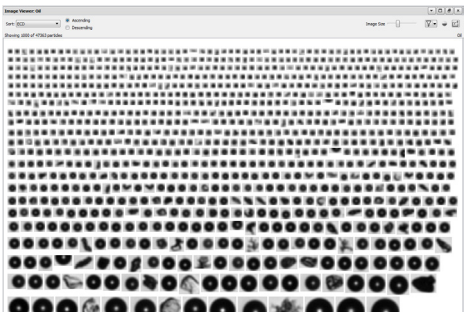
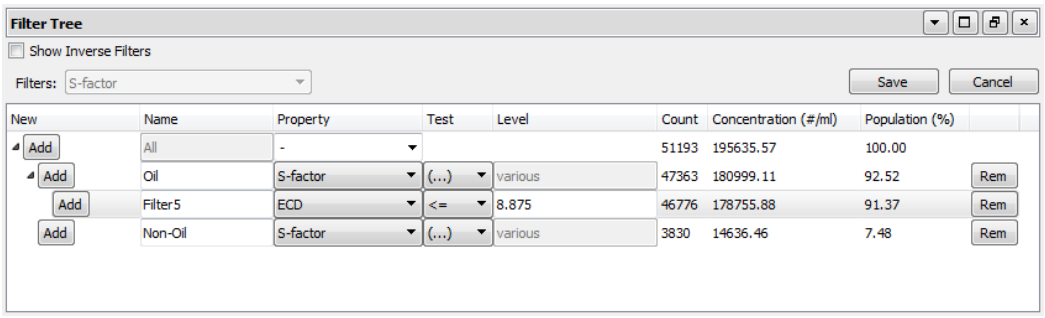


FIGURE 1. Image Analysis software makes it easy to get to all the data you need to characterize your sample and the particles in it, by having all the data in one streamlined interface. Fully resizable and customizable interface (A); Expanded windows for better visualization of graphs and particle images (B, C).

FILTER TREE

The Filter Tree window allows you to easily set up and edit filters based on 10 different morphological parameters (**Figure 2A**). If you import a dataset from MFI View System Suite (MVSS), any filters set up during sample running and image acquisition will be brought over into Image Analysis Software for a more cohesive analysis workflow. The new software features also include built-in filters, such as USP 787 and S-Factor, and you can also easily look at inverse filters by simply checking the **Show Inverse Filters** box (**Figure 2B**).

A.



B.

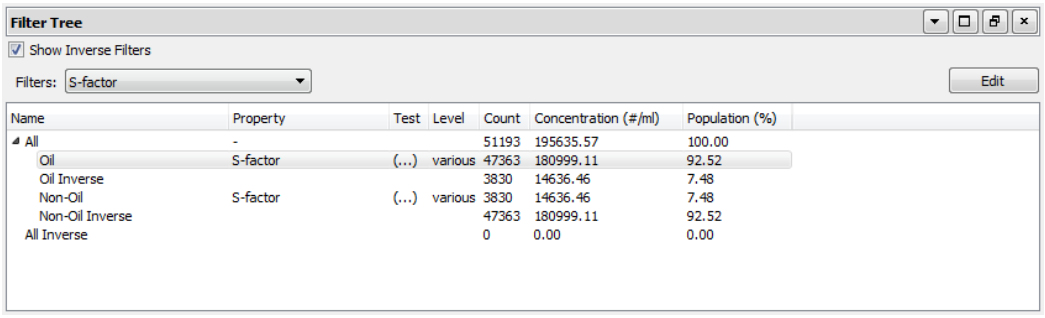


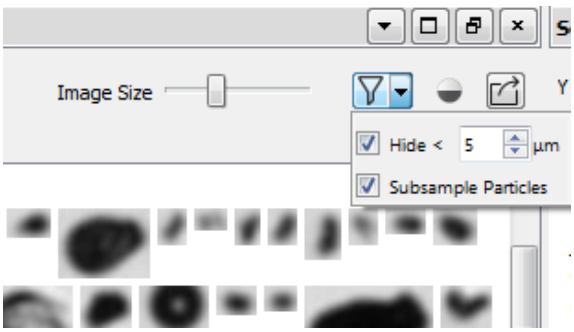
FIGURE 2. Using built-in filters. Editing the Filter Tree (A); Showing inverse filters (B).

IMAGE VIEWER

Image Analysis Software provides improved performance and fast image loading and viewing by giving you the option to view a subsample of the selected particle population. Want to see images of every particle? Simply uncheck the **Subsample Particles** box (**Figure 3A**).

Particles below a certain size can also be hidden and a slider bar lets you increase or decrease the size of images (**Figure 3A**). Want to sort images based on morphological parameters? No problem. The Image Viewer interface includes options for doing so in ascending or descending order (**Figure 3B**).

A.



B.

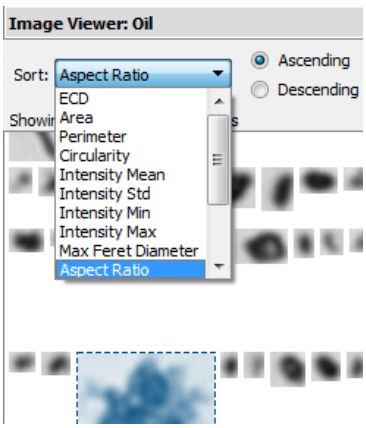


FIGURE 3. Controls in Image Viewer. Adjust the image size using the slider bar; also shown are subsamples of particles and those hidden based on size (A). Sort particle images based on morphological parameters using the Sort dropdown menu (B).

Hovering over a particular particle image in the Image Viewer will display a list of morphological data for that particle. (**Figure 4**).

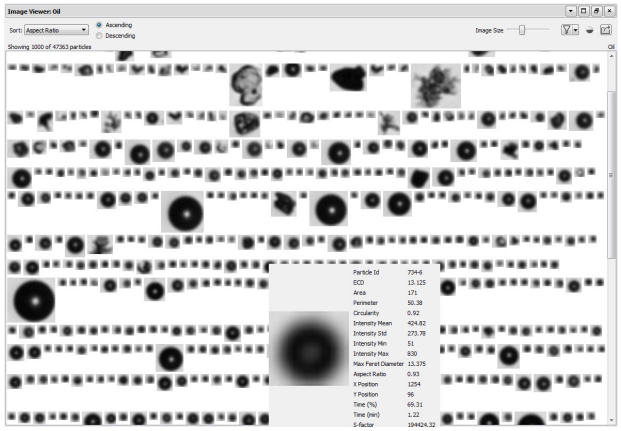


FIGURE 4. Hover over a particle in the Image Viewer to display morphological data.

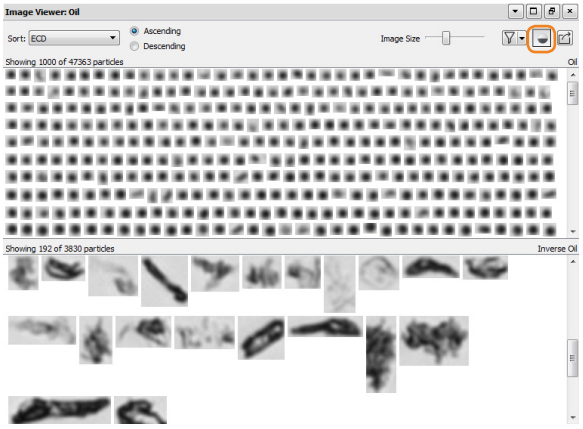
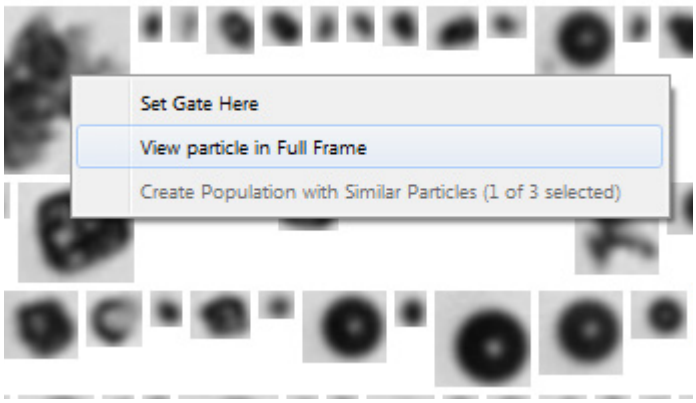


FIGURE 5. Show Inverse Population data display example.

If you've selected a filtered population, clicking the **Show Inverse Population** button in the top right corner of the view window will display particle images that are outside of the selected filter (**Figure 5**). For example, selecting the **Show Inverse Population** function for an applied filter pertaining to silicon oil particles would display non-silicon oil particles in the panel below.

And finally, for an expanded view of a particle of interest, the Image Viewer lets you view the selected particle within the full image frame (**Figure 6**). To do this, left-click on the particle of interest to select it, then right-click and select "**View particle in Full Frame**."

A.



B.

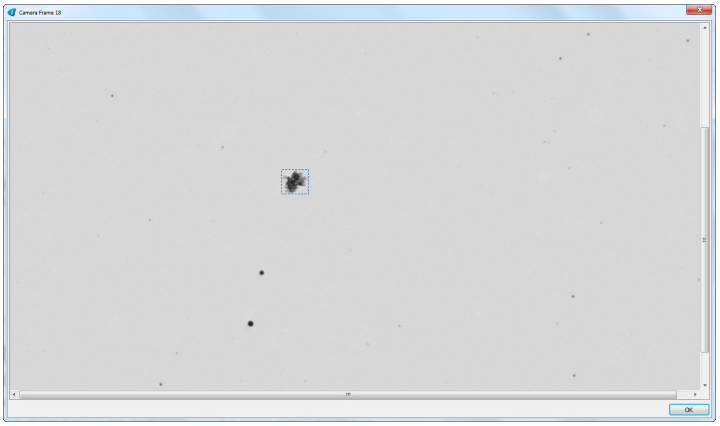


FIGURE 6. Expanded particle view. Select a particle of interest (A) to obtain a Full Frame image view (B).

GRAPHING TOOLS

To further up your data analysis game, you can create scatter plots to assess the relationship between different morphological parameters (**Figure 7A**) or histograms with linear or log scales for particle count and concentration analyses (**Figure 7B**). The graphs can also be exported as .png files.

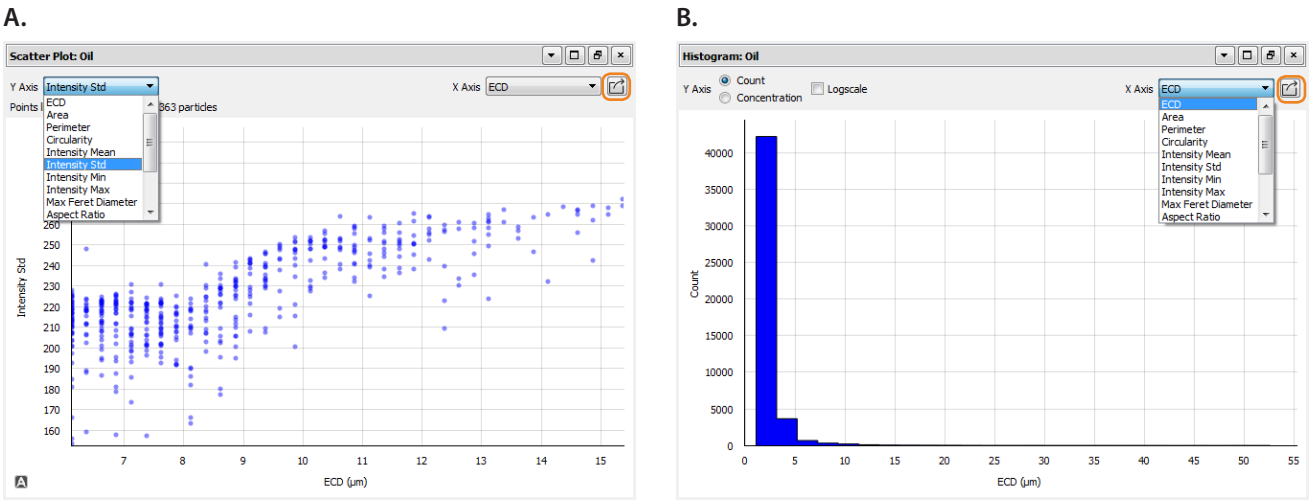


FIGURE 7. Graphing tools. Scatter plot comparing two morphological parameters (A). Histogram displaying count or concentration based on a morphological parameter (B). A graph image can also be exported as a .png file by clicking the arrow button in the top right corner of the user interface.

Analysis Tools

GATING FUNCTION

Image Analysis software makes it easy to define gates based on a single particle or a population subset from a graph. Sort your particle images, scatter plots, or histograms by your morphological parameter(s) of interest, and select the particle or subpopulation you want to gate on (**Figure 8**).

DYNAMIC STATISTICS

The Population Statistics window in Image Analysis Software allows you to quickly and visually see the all statistics for a selected population, like the average equivalent circular diameter (ECD) of a particle or the standard deviation for circularity. If you select a different particle population, the statistics window will update dynamically.

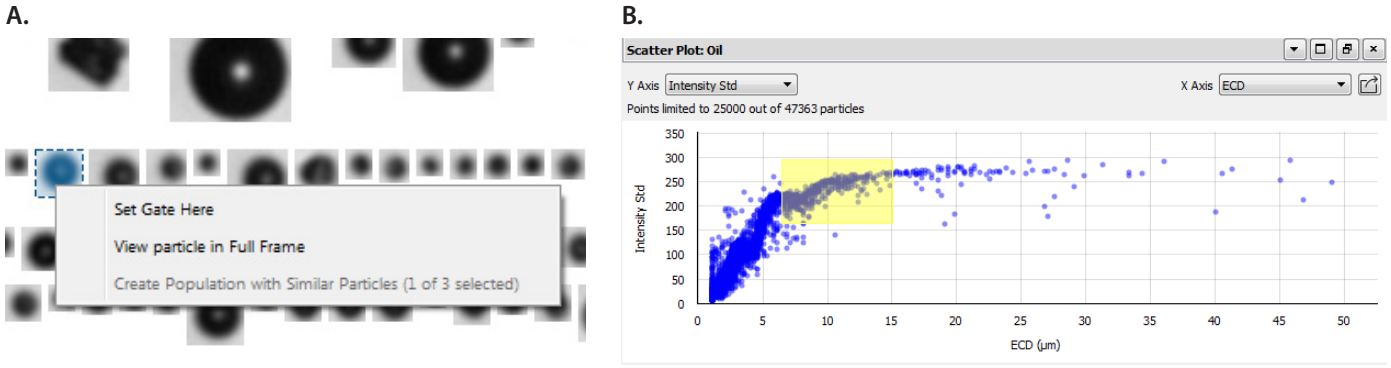


FIGURE 8. Setting a gate on a single particle (A) and selecting a subset on a graph to set a Local Zoom population (B).

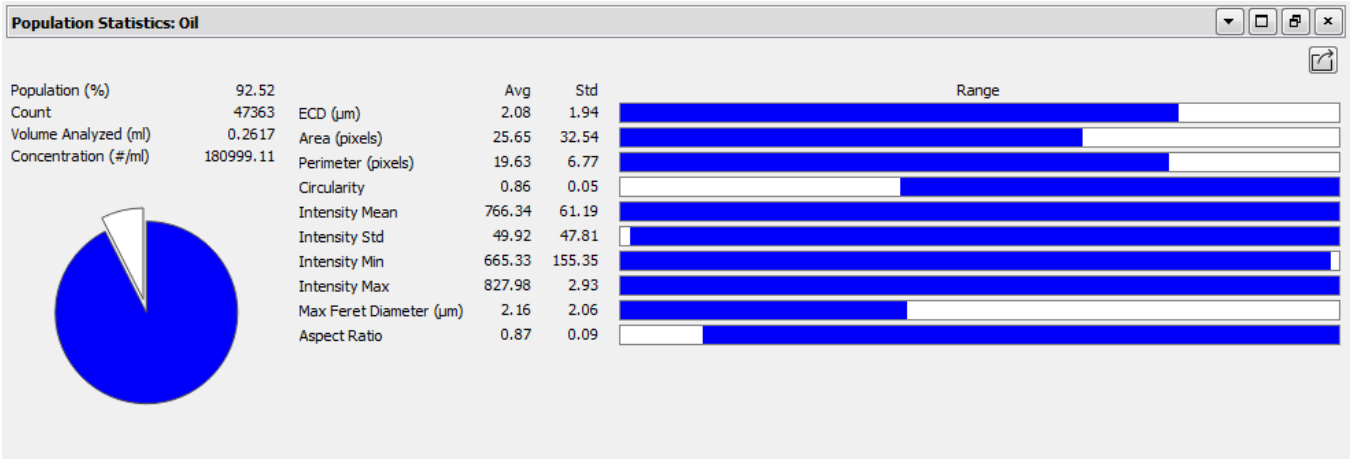
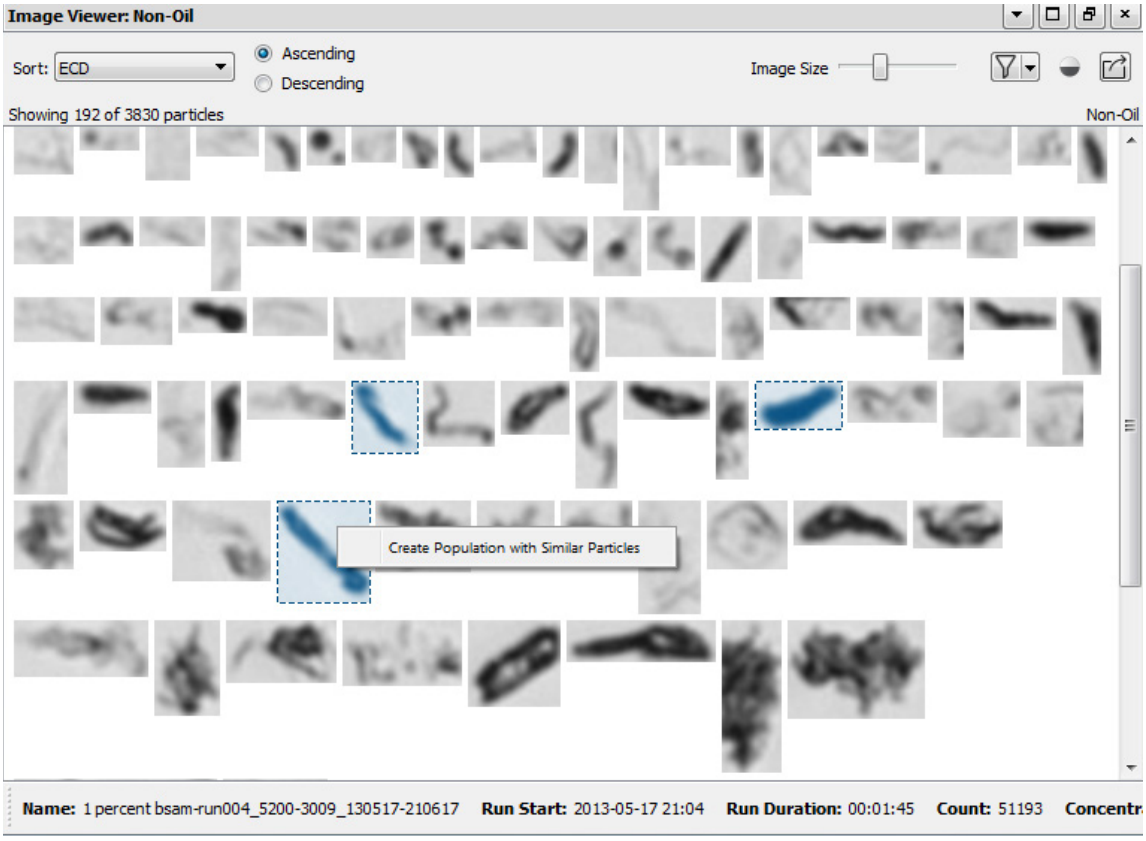


FIGURE 9. The Population Statistics window updates dynamically based on the selected particle population.

SEE SIMILAR PARTICLES

Image Analysis Software also has a powerful tool that makes it easy to filter out particles that are similar to each other. Select three or more particles in the Image Viewer, right-click, and select **Create Population with Similar Particles** (**Figure 10A**). Image Analysis will create new filters based on the morphological characteristics of the particles you selected. These filters can be viewed in the Filter Tree (**Figure 10B**) and saved if desired. The Image Viewer will display the particles that fall within this new filter (**Figure 10B**).

A.



B.

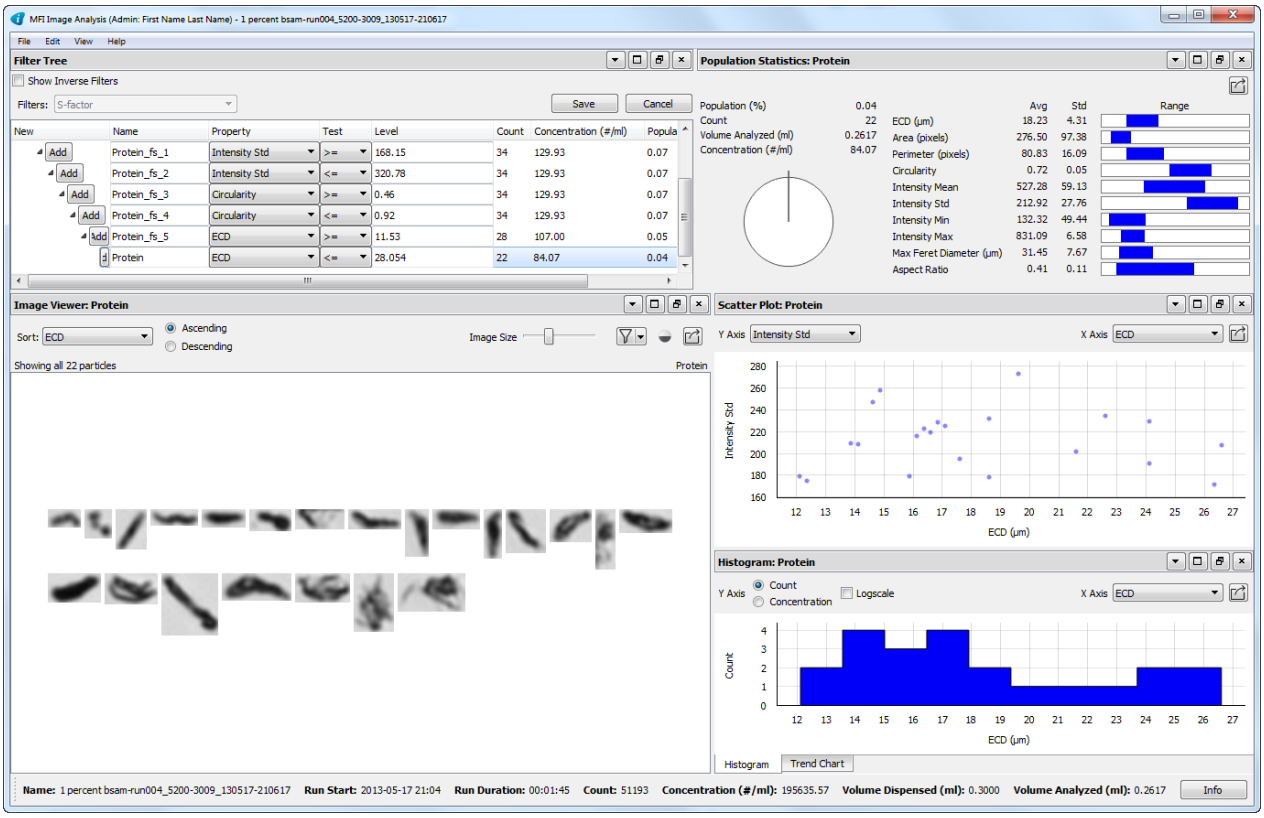
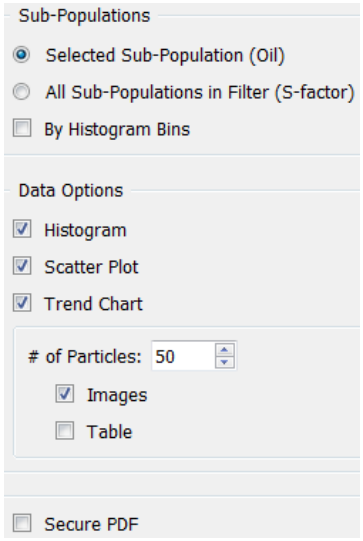


FIGURE 10. Find similar particles. Selecting particles and creating a population of similar particles (A). Updated display with new filters and images of similar particles (B).

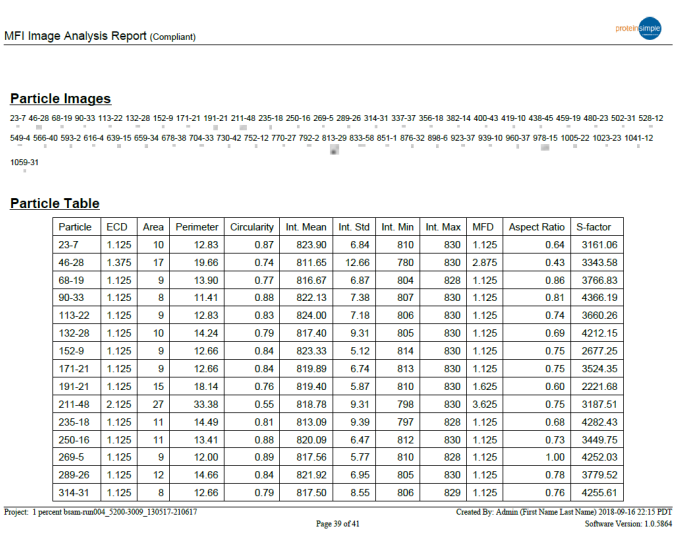
Customized Reports

The reports you generate can include as many particles per size bin as you see fit; just check off what you need in the **Create Sample Report** dialog box and how you'd like the data to be displayed (**Figure 11A**). The software can now generate charts, images and particle tables for varying size bins in the same sample (**Figure 11B**). When you're done, protect your information by saving the report as a secure, password-encrypted PDF that no one can alter.

A.



B.



Particle	ECD	Area	Perimeter	Circularity	Int. Mean	Int. Std	Int. Min	Int. Max	MFD	Aspect Ratio	S-factor
23-7	1.125	10	12.83	0.87	823.90	6.84	810	830	1.125	0.64	3161.06
46-28	1.375	17	19.66	0.74	811.65	12.66	780	830	2.875	0.43	3343.58
68-19	1.125	9	13.90	0.77	816.67	6.87	804	828	1.125	0.86	3766.83
90-33	1.125	8	11.41	0.88	822.13	7.38	807	830	1.125	0.81	4366.19
113-22	1.125	9	12.83	0.83	824.00	7.18	806	830	1.125	0.74	3660.26
132-28	1.125	10	14.24	0.79	817.40	9.31	805	830	1.125	0.69	4212.15
152-9	1.125	9	12.66	0.84	823.33	5.12	814	830	1.125	0.75	2677.25
171-21	1.125	9	12.66	0.84	819.89	6.74	813	830	1.125	0.75	3524.35
191-21	1.125	15	18.14	0.76	819.40	5.87	810	830	1.625	0.60	2221.68
211-48	2.125	27	33.38	0.55	818.78	9.31	798	830	3.625	0.75	3187.51
235-18	1.125	11	14.49	0.81	813.09	9.39	797	828	1.125	0.68	4282.43
250-16	1.125	11	13.41	0.88	820.09	6.47	812	830	1.125	0.73	3449.75
269-5	1.125	9	12.00	0.89	817.56	5.77	810	828	1.125	1.00	4252.03
289-26	1.125	12	14.66	0.84	821.92	6.95	805	830	1.125	0.78	3779.52
314-31	1.125	8	12.66	0.79	817.50	8.55	806	829	1.125	0.76	4255.61

FIGURE 11. Creating customized reports. Dialog box for configuring a report, including the option to save as a secure PDF (A). An example table from a report (B).

21 CFR Part 11 Compliant Analysis

MFI Image Analysis Software can now directly integrate with MVSS, to give you a streamlined, compliant workflow, when doing analysis on the instrument computer (**Figure 12**). Image Analysis software can directly access the repository, allowing you to analyze images from CFR-compliant files.

Actions performed in Image Analysis software on the instrument computer are logged in the audit trail and recorded for compliance.

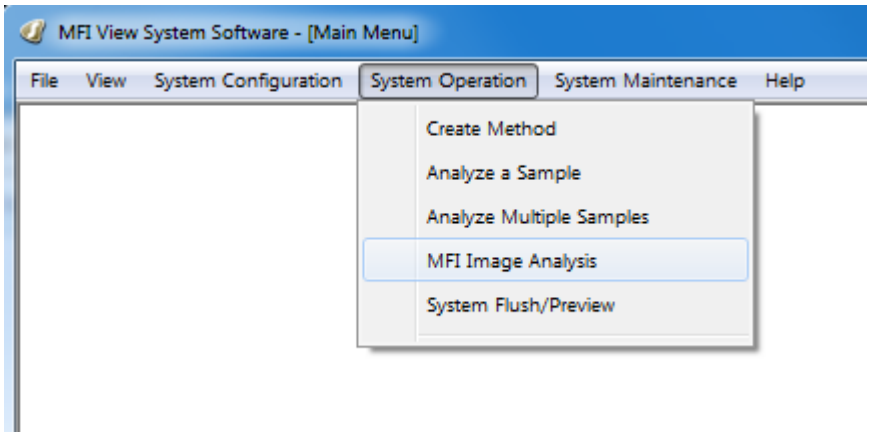


FIGURE 12. Launch MFI Image Analysis Software directly from MVSS.

Conclusion

MFI Image Analysis Software gives you the tools you need to analyze individual samples or cohorts of data according to the size, count and morphology of potentially present sub-visible particles in your product. In this technical note we've outlined various new features in the user interface that are sure to streamline your analysis and reporting workflow, which allow you to make the right decisions about the development and manufacture of your product.

proteinsimple
a bio-techne brand

Toll-free: (888) 607-9692
Tel: (408) 510-5500
info@proteinsimple.com
proteinsimple.com

PL7-0024 Rev A